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Building a Better Soil for Upland Surface Mine Reclamation in Northern Alberta: Admixing Peat, Subsoil and Peat Biochar in a Greenhouse Study with Aspen

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Abstract

Surface mining of oil sands in northeastern Alberta is a large-scale disturbance affecting over 900 km^2 so far. Extraction companies are required by law to return the environment to ‘equivalent land capability’, but this has been challenging to quantify. To date, only one site has been certified as reclaimed. Restoring ecosystem function, including nutrient availability and uptake, might be a more realistic goal of reclamation. We tested the effect of admixing subsoil with peat and peat biochar on bioavailable nutrients, foliar nutrient concentration, and aspen (*Populus tremuloides* Michx.) productivity in a greenhouse study. Brunisols and Luvisols, found in upland boreal forests of the Athabasca Oil Sands Region, have higher mineral soil content compared to the commonly used peat. Charcoal is a native component of boreal forest soils in northern Alberta and affects a variety of soil characteristics. In two separate tests, we compared different peat-subsoil admixtures, and biochar amended peat-subsoil admixtures to forest-floor-mineral-mix. Seedling productivity increased with admixing subsoil in both experiments with and without biochar, and there was an overall positive effect of amendment with biochar when comparing all treatments of both experiments using multivariate statistics, with biochar being more similar to FFM. Our findings suggested that peat-subsoil mixes did not provide sufficient amounts of P and Cu to seedlings. A lower K and Mn availability in peat-subsoil mixes was also identified and needs to be evaluated in further studies.

Keywords: biochar, land reclamation, soil prescriptions, plant nutrition, Populus tremuloides

INTRODUCTION

Land reclamation following surface mining frequently results in poor ecosystem function due to a lack of organic matter (OM) in cover soils. This applies to oil and gas operations in Wyoming (Mason et al. 2011), coal extraction in Germany (Šourková et al. 2005), or Bauxite mining in Australia (Jasper 2007; Lewis et al. 2009). However, surface mining of oil sands in boreal mixed-wood forests of northern Alberta suffers from the opposite problem. Selectively salvaged peat from lowland ecosystems during pre-mining (Fung and Macyk 2000) produces an excess of OM. This salvaged peat is used to reclaim upland ecosystems as a means of stimulating soil fertility, but the functional parameters created by this practice are not similar to those found in natural upland ecosystems (MacKenzie and Quideau 2010, 2012; Quideau et al. 2013; Sorenson et al. 2011). In fact, reclaimed ecosystems using peat are potentially put on recovery trajectories that create novel ecosystems (Audet et al. 2014).

In Alberta, surface mining of oil sands creates a large-scale ecosystem disturbance requiring *"reclamation .... to its equivalent capability"* (Alberta 2014a: 99). Conservation and reclamation regulations for the province of Alberta currently define equivalent capability as the ability to support various land uses, similar to what existed prior to surface mining, but not necessarily identical (Alberta 2014b). Legislation also requires evaluation of physical, chemical and biological characteristics in reclaimed ecosystems, including soils and vegetation following reclamation (Alberta 2014b). Together these two points imply re-establishment of ecosystem function on both local and regional scales. Of the 904 km^2 disturbed to date, only 104 ha of the total disturbed area has been certified reclaimed (Environment & Parks 2015). Another 5,901 ha have been permanently reclaimed, but must be monitored for more than 15 years before being certified (Alberta 2013). Currently, the presence and growth of local plant species are used to assess recovery of boreal forests on these disturbed areas (Alberta 2013; Environment&Parks 2015) along with concepts of fertility that may not be realistic for wildland ecosystems. Upland reclamation of surface mines in the AOSR is carried out by placing coversoil, mostly peat, over suitable subsoil or overburden. Operators are legally required to salvage all available topsoil (Alberta Environment 2004), which includes limited salvaged material from uplands that contain native seed and propagules. This material, when using it for reclamation, is called forest floor mineral mix (FFM). While vegetation recovery using FFM or LFH donor material as cover soil is better for reclamation (Mackenzie and Naeth 2010), the material is scarce and current reclamation practices use peat instead on sites targeting boreal upland forest recovery.

To date, reclamation practices in the oil sands mining-affected areas of northeastern Alberta have not considered the influence of wildfire as part of a possible recovery strategy. The intensity, type, and prevalence of fire in these boreal ecosystems influences vegetation and biogeochemical cycling (Maynard et al. 2014), and structures Western Canada's boreal mixed woods (Cumming 2001), as fire is the main natural disturbance of these ecosystems (Cumming 2001; Lecomte et al. 2006). It thermally alters organic matter, turning vegetation into charcoal, a stable carbon compound and a long-term carbon sink (Ohlson et al. 2009). Charcoal is a residue of wildfire (Preston 2009), and native vegetation might be as it is adapted to fire as a natural disturbance, be adapted to the influence of charcoal as a soil component, but this is not known to date. Biochar is a man-made surrogate for charcoal and can have the same influence on soil biogeochemical properties. Biochar has been used as a soil amendment for forest restoration since the early 1820's and is known to increase tree biomass (Thomas and Gale 2015). The addition of biochar to peat in greenhouse trials increased plant biomass production and reduced decomposition rates (Tian et al. 2012). Thomas and Gale (2015) suggested that biochar could potentially replace other forms of organic matter or liming agents in the field of forest restoration. Biochar might also stabilize carbon, change biogeochemical characteristics, influence nutrient mineralization, enhance plant performance, reduce greenhouse emissions, and increase soil microbial biomass in reclaimed soils (Anderson et al. 2011).

A possible solution to the issue of adding too much organic matter to reclamation soils in the AOSR is admixing peat with non-saline/sodic sub-soil material to create a soil prescription similar to natural, undisturbed forest soils in the region. These natural soils have low surface organic matter content and can be coarse textured with rapid drainage (Fung and Macyk 2000). Reducing surface OM content through admixing might better simulate these properties. Another solution for reclamation could creating biochar using available peat sources and incorporating it into peat-subsoil admixtures. Biochar can be chemically similar to pyrogenic carbon produced after wildfire and has been shown to influence soil biogeochemical processes (DeLuca et al. 2009).

Our study included two parts. In part one, we assessed the effect of admixing peat with non-saline/sodic sub-soil material on aspen biomass production and nutrient availability to create a reclaimed soil more similar to FFM. In part two, we examined the effect of including pyrolysed peat (biochar) as a Pyrogenic Carbon (PyC) amendment to admixed soils on aspen biomass production and nutrient availability, to determine if this produced soil more similar to FFM.

METHODS

Greenhouse Set-up

A greenhouse experiment was set up in March 2015 at the plant growth facility located at the University of Alberta, Edmonton, Alberta, Canada. Aspen *(Populus tremuloides* Michx.*)* seeds were germinated and grown for six weeks in styroblock containers (Beaver Plastics ID Code 540/8) and filled with Sunshine® Peat Moss. An 8ml container size was chosen to reduce the transfer of nutrients into reclamation soil types. All seedlings were fertilized two weeks after germination with 11-41-8 (NPK) tree seedling fertilizer (Agrium Advanced Solutions) according to manufacturer instructions. In mid-April 2015, seedlings were transferred into pots containing different admixed soil treatments (see below). Only seedlings showing equivalent growth were selected for transfer into treatment pots. Seedlings were watered daily to field capacity. Additional lighting was set to 16 h per day (6:00-22:00 hours).

Pots used for the trial were 20 cm in diameter and 10 cm in height for a total of volume 2.42 l. Pots were filled with one of six different admixtures of peat and sub-soil salvaged from the pre-mining environment. Peat was salvaged from lowland ecosystems and had a woody nature, consistent with paluaudification, while sub-soil was salvaged from a coarse-textured jack pine (*Pinus banksiana* Lamb.)

ecosystem. Admixtures were produced by mixing large quantities (40 l total per run) of peat and sub-soil in a clean cement mixer in different volumetric proportions for 10 minutes. FFM was mixed following the same procedure and consisted of the LFH and the top 10-15 cm of A/B horizon from the same coarse textured a or b ecosites. Type a ecosites are characterized by xeric to subxeric moisture regimes with poor to very poor nutrient regimes, while type b ecosites are characterized by submesic to subxeric moisture regimes with medium to poor nutrient regimes (Beckingham and Archibald 1996). Each experiment (admixing and admixing with biochar) was replicated six times with trees and three times without trees (Table 1). The experimental set up was a completely randomized design and pots were rotated in the greenhouse weekly.

Biochar was produced by gradual carbonization of peat in a muffle furnace in a reduced oxygen environment (tinfoil and sand bed). Peat was charred by fast-ramping (30 minutes) to a temperature of 500°C where it was held for one hour. Biochar was of fine structure with a BET (Brunauer–Emmett–Teller) surface area of 23 m^2 g^{-1}, total carbon content of 23.58% (C), hydrogen content of 0.78% (H), nitrogen content of 0.51 % (N), and oxygen content of 5.78 % (O). Each pot received 32.2 g of biochar, equivalent to 10 MT biochar/ha, mixed into the treatment pots. Treatment pots were incubated in the greenhouse for two weeks before trees were transferred into individual pots. Pots were watered with deionized water during the incubation period, to maintain 80 % Field Capacity.

Biomass and Laboratory Analysis

We measured bioavailable nutrients by installing Plant-Root-Simulator™ probes (PRS) in pots without trees according to the manufacturer instructions (Western AG Innovations, Saskatoon, SK). PRS™ probes employ membrane bound ionic exchange resins to capture nutrient anions and cations available in soil solution. Probes were incubated in-situ for one week prior to the end of the experiment. Upon removal from the pots, probes were rinsed with de-ionized water, sent to Western Ag Innovations, extracted with 0.5 M HCl and analyzed by colorimetry (FIAlab 2600) for NH_4^+ and NO_3^-, and by ICP-OES (Perkin Elmer ICP-OES 8300) for Al, B, Cu, P, K, Mn, N, Zn, Fe, Mg, Ca, and S.

The experiment ended in mid-July 2015 at which point the trees had been growing in the experimental treatments for 12 weeks. Roots were washed, and soil samples were collected. Plants were dried to a consistent weight at 65°C and total biomass of each tree was recorded. Leaves were ground up using a Ball Mill MM200 (Brinkmann Retsch). Elemental analysis of foliar material was conducted after its digestion with 65% HNO_3 in a MARS 5 microwave digester (CEM corporation). Foliar C and N were measured by dry combustion (COSTECH 4010 Elemental Analyser) and Al, B, Cu, P, K, Mn, Zn, Fe, Mg, Ca, Mo, Ni, Co, Na, Cr, and S were measured using ICP-OES (ThermoFischer iCap 6300 Duo). Elemental concentrations were controlled and verified with standard reference materials.

Soil samples were dried at 60°C to a consistent weight. All samples were sieved to remove plant debris, rocks and other soil components greater than 2.0 mm and then ground using a Ball Mill MM200 (Brinkmann Retsch). Samples were transferred to 20-mL scintillation vials and stored before analysis.

Soil thermal stability was measured on twenty percent of the samples using Differential Scanning Calorimetry (DSC; STA 6000, Perkin Elmer). Approximately 20 mg of each sample were loaded in an open ceramic pan under an oxidative atmosphere (flow rate: 20 mL min^{-1} Oxygen gas and 80 mL min^{-1} Nitrogen gas) at a scanning rate of 20°C min^{-1}. The heat of combustion (Q in J g^{-1}) was determined by integrating the DSC curve over the exothermic region (150°-550°C). The area on the DSC curve was divided into three groups based on resistance to oxidation: (1) labile organic matter (150°C-375°C), (2) recalcitrant organic matter (375°C-475°C) and (3) highly-recalcitrant organic matter (475°C-550°C) modified from Merino et al. (2014), Merino et al. (2015), and Merino et al. (2016). These partial heats of combustions were designated as $Q1$, $Q2$ and $Q3$, respectively following Merino et al. (2014), for convenience. Soil thermal stability was calculated as a ratio of recalcitrant ($Q2$) over the labile organic matter ($Q1$).

Diffuse Reflectance Infrared Fourier Transform Spectroscopy (DRIFTS) was performed on all samples to predict soil thermal stability measured by DSC (Merino et al. 2015). Samples were placed in stainless steel sample cups and loaded into a Cary 600 Series Fourier Transform Infrared Spectrophotometer (FTIR, Agilent Technologies) equipped with diffuse reflectance accessory

(AutoDiffusIR^{TM}, PIKE Technologies). IR spectra of each soil sample were generated based on 16 sample scans per run at a spectral resolution of 4.0 cm^{-1}. All samples were analyzed in triplicate. IR spectra were baseline corrected and averaged before partial least square regression analysis (PLSR).

PLSR analysis of the prediction of heats of combustion from FTIR spectral data was performed using Unscrambler X (CAMO Software AS ver.10.3). The heat of combustion from labile organic matter (*Q1*) was predicted from IR spectral ranges of 1000 cm^{-1} - 1300 cm^{-1} (-C-H bending and C-O stretching), 2850 cm^{-1} - 2950 cm^{-1} (alkyl C-H stretching) and 3380 cm^{-1} - 3410 cm^{-1} (O-H stretching). The heat of combustion from recalcitrant organic matter (*Q2*) was predicted on the regions for unsaturated bond vibrations: 750 cm^{-1} - 850 cm^{-1} (=C-H bending) and 1500 cm^{-1} - 1850 cm^{-1} (C=C stretching and C=O stretching).

Statistical Analysis

R 2.1.5 (R Core Team 2012) was used for statistical analyses with *agricolae* package (de Mendiburu 2014) for post-hoc testing with *Tukey HSD* test and with permutational ANOVA in the *lmPerm* package (Wheeler 2010) for significance testing. Biochar amended treatments were tested separately within another set of the admixed soil treatments. Following separate analysis, all treatments were compared by Principal component analysis (PCA) using the *ggbiplot* package (Vu 2011) in R3.3.1 (R Core Team 2016). Data were standardized before PCA by log transformation and adding a constant (log10(x+100)).

RESULTS

Substrate Characterisation

Values for pH ranged from 6.8 in FFM to a high of 8.0 in S100 and P30S70 soils with biochar added in both. Treatments with peat concentrations >50% had pH values ranging from 7.6 to 7.9. Values for pH were higher in soils with biochar than in soils without biochar (Table 1) while pH of FFM soils was lower than all other treatment soils. EC values in all soil mixes with biochar ranged from 292 to 547µs/cm and were higher than the EC of FFM (161 µs/cm). EC values of all soils without biochar were also higher than FFM except for S100 (87µs/cm) (Table 1). Organic matter (OM) content values were higher in soil mixes with biochar than without biochar except for P80S20 (Table 1). OM content for FFM soils was 4.1%, slightly higher than S100 and P30S70 values regardless of biochar being present but substantially lower than those soil mixtures with higher percentages of peat (Table 1). Total Carbon (TC) and Total Nitrogen (TN) were higher in treatments with more peat. Biochar amendment increased TC and TN compared to unamended treatments (Table 1).

Effect of Admixing Peat

Biometrics data

Biomass of seedlings grown in FFM was significantly greater than seedling biomass of all other treatments except for P50S50. Biomass of seedlings grown in P80S20, P90S10 and P100 were significantly lower than FFM and P50S50 (Table 2). Seedlings grown in FFM were significantly taller than all other treatments and almost 90% taller than the best performing admixture P50S50 (Table 2).

Foliar data

Mean foliar concentrations of N were significantly higher in P80S20, P90S10 and P100 than all other treatments including FFM (Table 2). Concentrations of P were significantly greater in FFM and S100 than all other treatments and up to 300% greater than in treatments with >80% peat admix ratio. Mean values of P in P30S70 were also significantly greater than P80S20, P90S10 and P100. Foliar concentrations of K were significantly higher in P90S10 than P30S70 and S100 (Table 2). Mean K concentration from FFM was only significantly greater than K concentrations in S100 (Table 2). Average foliar S concentration was significantly higher (100% greater) in P80S20 than P50S50, P30S70, S100 and FFM (Table 2). Foliar Mg concentrations were significantly higher in S100 than all other treatments including FFM (Table 2). Foliar Ca concentrations were significantly higher in P30S70 than P90S10 and P100 (Table 2). Foliar Na concentrations were significantly higher in P90S10, and P80S20 than P50S50, P30S70 and FFM, the foliar concentration of Na was up to 600% greater in treatments with high peat admix ratios (Table 2). Foliar Mn concentrations were significantly higher in S100 than in FFM and P30S70 and all three treatments were significantly higher than all admix treatments with peat (Table 2). Foliar Cu concentrations were highest in FFM but not significantly higher than in S100, P80S20, P90S10, and P100 (Table 2). Cu concentrations in the foliage from P50S50 and P30S70 treatments were less than 50% of concentrations in FFM seedlings and significantly different to FFM with means below 5 $\mu g\ g^{-1}$.

The axes of principal component (PC) 1 and PC 2 of standardized foliar nutrient concentrations from aspen seedlings explained 50% of the variation in data (Figure 1). The axis of PC1 showed the influence of peat and subsoil proportions. FFM samples grouped midway between all peat (P100) samples on the left and all subsoil (S100) samples on the right. As peat concentration decreased, groups moved to the right with samples containing P50S50 and P30S70 overlapping with FFM samples. Higher foliar concentrations of P, Mn, Co and Mg were found in S100 samples. Foliar K and N concentrations were positively related to P100 samples while P was negatively related to samples containing more peat. The axis of PC 2 showed higher foliar Na and S concentrations that were negatively related to FFM samples. Analyses of PC 3 and PC 4 (data not shown) increased total explained variation to 73.3 percent but could not identify significant differences among peat treatments. However, this analysis highlighted differences in foliar Zn, Mn, P, and Cu concentrations in FFM from other treatments. Foliar concentrations of Zn were not significantly different among treatments when tested separately.

PRS and SOM stability data

Bioavailability of N, S, Mg and Ca were significantly higher in P80S20, P90S10 and P100 treatments than P50S50, P30S70, S100 and FFM treatments. Bioavailability of P was significantly higher in FFM and exceeded levels of other treatments by 400%. Bioavailability of K was also significantly higher in FFM than in all other treatments except for P90S10. SOM stability was significantly greater in P80S20 than in P50S50, P30S70, S100 and FFM.

PCA axes PC1 and PC2 of standardized bioavailable nutrients and soil organic matter stability explained 70.1% of the variance in data (Figure 2). As in Figure 1, PC1 axis in Figure 2 showed the influence of peat and subsoil proportions with FFM samples grouped midway between all peat (P100) samples on the left and all subsoil (S100) samples on the right. As peat content decreased, and subsoil content increased, groups moved to the right with samples containing P50S50 and P30S70 in line with FFM samples. Axis PC2 in Figure 2 showed a separation between FFM samples and all admixed soil samples which suggests differences between natural soils (FFM) and the admixed soils created for this study. Bioavailability of P was positively related to FFM while most other nutrients were positively related to peat content and negatively related to subsoil content. Figure 2 clearly showed treatment P50S50 had a nutrient profile similar to FFM with regards to the variation explained by the axis of PC1. Differences between P50S50 and FFM related to the differences of bioavailable P, Mn, K, and Cu on Axis PC2.

Effect of Admixing Biochar to reclamation soil mixes

Biometrics data

Biomass of seedlings grown in FFM was significantly greater than the biomass of seedlings grown in all other soils mixtures with biochar except P50S50+ which, on average, produced only 55% as much biomass as seedlings grown on FFM. Biomass of seedlings in P50S50+ did not differ significantly from all other admixture treatments with biochar (Table 3). Seedling height followed the same pattern as seedling biomass. Seedlings in FFM grew significantly taller than all other admixtures with biochar during the study except seedlings in P80S20+ with biochar; however, seedlings grown on FFM were more than 70% taller than the best performing admixture. Seedling height in P80S20+ did not differ significantly from any of the seedlings grown in other soil admixtures with biochar (Table 3).

Foliar data

Foliar N concentration in FFM seedlings did not differ significantly from any other soil treatments with biochar except S100+. Foliar N concentration was significantly greater in P100+ than P50S50+, P30S70+, and S100+ (Table 3). Foliar P concentration was significantly greater in FFM seedlings than all other soil admixing treatments with biochar except for P50S50+. P concentration in P50S50+ was significantly greater than P80S20+, P90S10+ and P100+ (Table 3). Foliar K concentration was significantly higher in P80S20+ and P90S10+ than in S100+, while foliar concentrations of K from FFM did not differ significantly from any seedlings of the treatments with biochar. Foliar concentrations of S and Mg did not differ significantly among admixtures with biochar or from FFM (Table 3). Foliar concentrations of Ca were significantly greater in P50S50+ than all other soil admixing treatments with biochar including FFM. Foliar concentrations of Na in FFM and P50S50+ were significantly lower than S100+, but there were no significant differences in Na concentrations among any of the other soil admixing treatments. However, foliar Na concentrations were 100% to 600% greater than in FFM (Table 3). Foliar Mn concentrations were significantly smaller in all peat subsoil admix treatments compared to FFM. Foliar Mn concentrations from FFM were more than 100% greater (Table 3). Foliar Cu concentrations were higher on FFM compared to all treatments. However the difference was significant in all cases (Table 3). Treatments P80S20+, P50S50+, and P30S70+ were significantly different to FFM in foliar Cu concentration with mean Cu concentrations below 5µg g^{-1} (Table 3).

PRS available nutrients and SOM stability data

N bioavailability, determined using PRS™ probes, was significantly higher in P100+ than in P50S50+, P30S70+, S100+, and FFM (Table 3). Bioavailability of P and K were significantly higher in FFM than all other soil treatment samples with biochar. Bioavailability of S did not differ significantly among any of the samples. Mg bioavailability was significantly higher in P80S20+ than FFM and S100+. Ca bioavailability was significantly greater in P80S20+ and P90S10+ than in P30S70+, FFM and S100+ (Table 3). SOM stability in treatments P100+, P90S10+, and P80S20+ was significantly greater than in P50S50+ and FFM. SOM stability was significantly lower in P30S70+ and S100+ (Table 3).

Biochar effect on admixing treatments

Permutational two-way ANOVA could not identify significant differences when comparing peat admix treatments with and without peat biochar amendment. For that reason the entire study was analysed using multivariate statistics (PCA) in order to capture the overall effect of a biochar amendment.

Figure 3 shows PCA results for all data together, including available nutrients, SOM stability, foliar nutrient concentrations, and biometrics (height, biomass). Axes PC1 and PC2 explained 53 % variation. The ordination showed a trend that with biochar amendment the treatments P100, P90S10, P80S20, P30S70, and S100 shifted closer towards FFM, suggesting overall characteristics of those treatments were similar to the reference substrate FFM (see arrows in figure 3). Treatment P50S50+ was less similar to FFM. Analysis of PC3 and PC4 for this PCA (data not shown) increased the total explained variation to 71.6 %, but did not identify trends or significant differences with biochar amendment.

DISCUSSION

Admixing

Improving growth

Aspen seedling biomass was greatest in P50S50 and did not differ significantly from aspen growth in FFM. Compared to the classical placement of reclamation materials in layers in upland oil sands mine reclamation (e.g. 30 cm of peat over 120 cm subsoil (Barber et al. 2015)), reclamation might be more successful with those materials by mixing materials to create conditions similar to natural soils. However, height growth in our admixture treatments was still significantly less than FFM soil. Height growth has a strong influence on competitive success, especially when competing for light in a weedy environment (Weinig 2000). Davis et al. (1999) reported that seedling survival was significantly affected by interactions of both water and light availability. Considering the increased height and biomass in P50S50, relative to the other admix treatments, survival rates of seedlings could be greatest on this cover soil prescription.

Assimilation of foliar nutrient concentrations to FFM grown seedlings

The foliar nutrient profile was influenced by admixing mineral subsoil and peat. Treatments P50S50 and P30S70 were most similar to FFM, with differences mainly explained by foliar S and Na concentrations (Figure 1). When all other treatments are compared to FFM, differences in foliar nutrient profiles can be interpreted as factors that reduced growth, even though in some cases, differences are not significant (Table 2). The shift in foliar nutrient profiles along axis PC1 (Figure 1) suggests that those treatments were significantly different. The driving elements on axis PC1 are P, Mn ,Co, Mg, Fe, Al, Cr, Cu, and Ca concentrations (Figure 1) which point towards treatment S100 and are negatively correlated with the foliar N and K concentrations related to treatments with a higher peat content as P100, P90S10, and P80S20. A more detailed analysis with permutational ANOVA and Tukey HSD revealed that concentrations of N, P, K, Mn, Cu, S, and Na were of greater importance (Table 2).

Kirkby (2012) stated that shoot dry matter concentration of N greater than 15 mg g^{-1} is required for adequate growth. All treatments, except S100, had foliar N concentrations greater than this critical value. With Foliar N concentration in peat ad-mix treatments higher than concentrations in FFM, which showed greater growth than the other treatments, N availability was not a limiting factor for aspen seedling growth in this experiment. Therefore, application of N fertilizer on peat cover soils for reclamation targeting type a or b ecosites is not needed, as supply rates of N are higher than on FFM, which is sourced from the targeted upland forest ecosystems.

Attiwill and Adams (1993) suggested that growth in forests is probably limited by P or some other elements, rather than N. The shoot concentration of P required for adequate growth is 2 mg g^{-1} (Kirkby 2012). None of the treatments reached that foliar P concentration, however, FFM and S100 had significantly higher foliar P concentrations than treatments with higher admix ratios of peat. For example, P100 had only 50 % as much foliar P concentration as P50S50 and significantly less accumulated biomass. P limitation can reduce shoot growth rate, inhibit formation of reproductive organs, or lead to restricted seed formation (Hawkesford et al. 2012). Those factors are critical for establishing a self-sustaining forest ecosystem. Our study showed that admixing increased foliar concentrations of P, however the increase was insufficient when foliar concentrations and growth of admix treatments were compared to FFM.

Foliar concentrations of K decreased with admixing subsoil to peat, but was similar to FFM up to a ratio of 50% subsoil. Foliar concentrations of K were greater than 10 mg g^{-1}, the value reported by Kirkby (2012) for adequate function. Only treatments P30S70 and S100 had K concentrations below 10 mg g^{-1}. K plays an important role in enzyme activation and osmoregulation (stomatal control), and K deficiency predisposes plants to abiotic and biotic stresses (Hawkesford et al. 2012). In the boreal forest ecosystems, the increased probability for plant damage under conditions of drought, or low temperature are important (Hawkesford et al. 2012). Massive drought-induced aspen mortality has been observed (Hogg et al. 2008; Michaelian et al. 2011), as well as thaw-freeze events inducing mortality, dieback, or reduced growth (Hogg et al. 2008).

Differences in foliar Mn concentrations could be importance since Mn deficiency is common in soils with high OM and pH (Broadley et al. 2012). The tested ad-mixes were all high in OM (peat) and are characterized by basic pH values ranging from 7.8 to 7.6 (Table 1). Only mineral subsoil (S100) and FFM had pH values slightly below pH 7. Admixing of subsoil lowered pH slightly which could potentially improve Mn availability of reclamation cover soils. The critical Mn value lies in between 10 and 20 $\mu g\ g^{-1}$ (Broadley et al. 2012), therefore a deficiency of Mn in our study was unlikely, but our data showed Mn concentrations lower in peat admix treatments than in FFM. However, our study was conducted over a short period (90 days). Future work needs to test if admixed cover soils can supply adequate Mn to support native vegetation over longer periods of time. Again, this is important for boreal reclamation, as Mn deficient plants are more susceptible to damage by freezing or soil born fungal root-rot diseases (Broadley et al. 2012).

Similarly to Mn, Cu deficiency has been described for soils that are high in OM (Broadley et al. 2012) and is related to the complexation of Cu with organic substances. Another factor that drives Cu deficiency is high N availability (Broadley et al. 2012). Values for critical Cu deficiencies range from 1-5 $\mu g\ g^{-1}$ depending on plant organ and species (Broadley et al. 2012). Typical symptoms of Cu deficiency include stunted growth, chlorosis/necrosis starting at the apical meristem, or bleaching of young leaves (‘white tip’ or ‘reclamation disease’) (Broadley et al. 2012), and similar symptoms were observed on seedlings in our study. Foliar concentrations were in the range of 5 $\mu g\ g^{-1}$ for the majority of admix treatments, but in P50S50 and P30S70 lower, so deficiencies potentially possible. Given the nature of reclamation soils being used, future work should evaluate potential Cu deficiencies in long-term field trials as N metabolism, lignification, and pollen formation are affected by Cu deficiency (Broadley et al. 2012).

Differences between P50S50 and FFM, and other treatments were also explained by axis PC2 (Figure 1). Foliar S and Na concentrations drive the variation between treatments along this axis. While S is a macro nutrient that can limit tree seedling growth (Ericsson 1995), Rennenberg (1984) suggested that excess S can negativly impact plant metabolism and can affect dry weight accumulation, yield, and delay flowering. With the short growing season in boreal climates and limited knowledge on the response of native vegetation to increased S concentrations, future studies should evaluate potential effects on flowering delay as this might be critical for reproduction of plants. Na is known to have potentially toxic effects on plants, even at low levels, and can negatively affect plant growth and development (Luan et al. 2009). Higher foliar Na concentrations were observed in all treatments relative to FFM. However, P50S50 had the lowest foliar Na concentration, which was not significant different from seedlings grown on FFM.

Assimilation of biogeochemical characteristics to FFM by admixing

Our study showed that admixing peat with mineral subsoil resulted in more similar biogeochemical characteristics with regard to nutrient availability and SOM stability, when compared to FFM. This is important when reclamation practices target plant communities that are adapted to specific nutrient regimes. When targeting type a/b ecosites, using a cover soil with high nutrient availability is inappropriate, as distinct plant communities occupy sites with low nutrient availability as a competitive strategy (Chapin et al. 1986). Results from our study suggest that nutrient availability in the admix treatment P50S50 is most similar to FFM based on the variation explained on axis PC1 (Figure 2) which is driven by decreased availability of P, Mn, K, and Cu. By fertilizing P50S50 with an adequate rate of those nutrients, very similar nutrient availability compared with FFM could be established. This would be of great importance for re-establishing native plant communities, as soil fertility is in many cases an excellent predictor for the composition of plant communities (Chapin et al. 1986; Christensen and Peet 1981). Future field studies should evaluate the effect of targeted fertilization on the establishment and resilience of native plant communities on P50S50 cover soil compared to equivalent ecosites following disturbance, and sites reclaimed using current standard procedures.

Amending biochar to admix treatments

Improving growth

Treatment P50S50+ showed the greatest biomass accumulation and did not differ significantly from FFM grown aspen seedlings, however, heights of seedlings in P50S50+ were significantly lower compared to FFM, which is critical when competing with other plants. Biedermann and Harpole (2013) showed that a biochar amendment can increase plant productivity by various mechanisms, however growth equivalent to FFM was not achieved. This might be the result of producing the biochar from peat (P100), which might be considered low quality feedstock given the low BET surface area of 23m^2 and a low C content. Properties of biochar are affected by feedstock quality and pyrolysis temperature (Kloss et al. 2012).

Assimilation of foliar nutrient concentrations to FFM

Foliar nutrient concentrations of N, K, S, Mg, and Na in biochar amended treatments P100+, P90S10+, P80S20+,P50S50+,P30S70+ were similar to foliar nutrient concentrations in FFM than in treatments without biochar. Foliar P concentrations, however were significantly different compared to FFM, indicating that the amendment of biochar did not improve P availability. Foliar P concentrations in biochar-amended treatments were still below critical values, indicating that the biochar used in this study did not stimulate uptake. Steiner et al. (2007) described a positive biochar effect on P uptake using secondary forest wood as a feedstock, however in our study probably not existing for P as the feedstock and other OM was very low in P. Similar observations were made for foliar concentration of Mn and Cu.

Biochar amendment increased pH and SOM content (Table 1) and subsequently reduced bioavailability of Mn and Cu. Biochar amendments reduced the fraction of exchangeable Cu and might have a negative effect on Cu availability to plants, as peat admix treatments had low Cu availability. Foliar symptoms indicating Cu deficiency were observed in biochar amended admix treatments. Foliar concentrations of N, P, K, S, Mg, and Na in P50S50+ did not differ significantly from FFM foliar concentrations, but Mn and Cu were lower. Significantly higher foliar concentrations of Ca in P50S50+ were within the range of 1,000 and 50,000 mg g^{-1} which was reported for plants depending on organs and growing conditions (Hawkesford et al. 2012) and likely had no adverse effects.

Assimilation of biogeochemical characteristics

Adding biochar produced similar N bioavailability among all treatments except in P100+. Biochar has increased bioavailability of NH_4-N shortly following application, but decreased NO_3-N concentrations consistently (Nelson et al. 2011). In our study, biochar amended treatments had a higher N availability than unamended treatments. When targeting a long-term status with lower N bioavailability that is comparable to FFM a biochar amendment might be useful, but that needs to be tested in the reclamation environment. None of the admix treatments amended with biochar reproduced similar P bioavailability. Nelson et al. (2011) stated that biochar can reduce the amount of exchangeable P when no P fertilization is applied. Since the bioavailability of P from the peat and subsoil used in our study was low, future studies should evaluate the effects of P fertilization with biochar amendment, or test biochars that are produced from feedstock that has a higher P content. Oram et al. (2014) showed that biochar can improve the availability of K to plants, but biochar did not elevate the availability of K to a level comparable with FFM in our study. For S bioavailability no significant differences with biochar amended treatments were identified, however the average bioavailability of S on treatments with higher peat contents (P100+ to P50S50+) was five times higher than FFM. Biochar releases essential elements like S (Uchimiya et al. 2010), however increased S availability needs to be evaluated carefully as excess S in plants can potentially have negative effects (Rennenberg 1984). Bioavailability of Ca and Mg in treatments P100+ to P30S70+ showed that admixing mineral subsoil and peat in combination with biochar amendment resulted in sufficient availability of those elements relative to FFM. SOM stability of P50S50+ was not significanty different from FFM. Stability of SOM can be used as a proxy to indicate how easily C and N can be mineralized (Plante et al. 2011). For this reason P50S50+ will provide a similar nutrient mineralization rate and availability as FFM. As the effects of biochar on the stabilisation of pre-existing SOM are not currently understood (Kimetu and Lehmann 2010), future work should examine the effect of biochar on peat, which is the surrogate of SOM in upland reclamation.

Evaluating the effect of peat biochar on peat subsoil admixtures using multivariate statistics

An analysis of all factors together, including available nutrients, foliar concentrations, biometric parameters, and biochar amendment (Figure 3) revealed that treatment P50S50 without biochar was most similar to FFM. However, biochar amendment shifted all other treatments towards FFM. This might be of great importance when there is a material shortage of peat or subsoil to recreate similar characteristics as found on FFM. The addition of biochar to peat cover soils seems promising, as it increases the similarity of overall biogeochemical characteristics and seedling performance to FFM. Research has shown that a biochar amendment can improve productivity of soils by affecting soil physical and chemical parameters (Ameloot et al. 2013). However, negative effects on productivity have been also reported (Ameloot et al. 2013). Future work should focus on evaluating the potential use of biochar in upland oil sands mine reclamation over longer periods of time in field settings and assess the impact of biochar derived from different feedstocks.

CONCLUSIONS

Admixing mineral subsoil with peat created cover soil characteristics similar to FFM. The best ratio in terms of tree growth, foliar nutrient, and available nutrient profiles was 1:1. However, foliar data indicated that P and Mn availability was insufficient and Cu was potentially deficient. Availabilities of those nutrients were not improved by admixing. PRS™ nutrient data also suggested that K availability was significantly lower on peat cover soils and subsoil compared to FFM. In regards to all other tested nutrients, a great similarity in foliar concentrations and bioavailability could be achieved with a 1:1 admix ratio. Concentrations P and Mn in biochar amended admix treatments were still too low , and Cu was also potentially deficient in various admix treatments with biochar amendment. In all admix treatments with and without biochar, PRS™ data suggested that K availability needs to be evaluated further. Biochar amendment increased the similarity of most peat admix treatments to FFM. This might be of importance when targeting plant communities adapted to certain soil characteristics or when an ideal admix ratio cannot be achieved due to availability of salvaged materials. Further studies should investigate the use of soil amendments that can additionally improve the availability of P, K, Mn, and Cu. We are suggesting to test local waste products (e.g. biosolids - as those are known to contain high amounts of e.g. Cu) or biochars produced out of materials rich in deficient elements to address differences in nutrient profiles preferably in line with a field study. Further research should also evaluate the influence of the addressed parameters on establishment of native boreal forest vegetation, ideally over several growing seasons. These studies should also assess recovery from natural disturbances, for example fire or experimental drought, to evaluate the long-term suitability of cover soil prescriptions in context of resistance and resiliency of targeted plant communities. Careful attention must be paid when applying greenhouse study results in the field. However, we are confident that considering our findings in land reclamation practice will be useful when aiming to re-establish a or b ecosites.

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Table 1: Experimental design and substrate characteristics.

Treatment	Peat/Subsoil ratio	Biochar	Material type	pH (aq)	EC (µs/cm)	%SOM (w/w)	%TC (w/w)	%TN (w/w)
P100	100/0	w/o	Peat	7.75 (0.04)	470 (64)	36.11 (0.03)	17.93 (0.14)	0.922 (0.119)
		w(+)		7.79 (0.16)	547 (146)	40.56 (0.05)	20.99 (0.02)	1.07 (0.13)
P90S10	88.9/11.1	w/o	Peat-Mineral-Mix	7.61 (0.04)	320 (27)	23.82 (0.04)	15.97	0.820
		w(+)		7.81 (0.03)	404 (56)	24.66 (0.64)	16.06	0.976
P80S20	80/20	w/o	Peat-Mineral-Mix	7.61 (0.02)	252 (323)	19.16 (0.07)	14.41	0.739
		w(+)		7.74 (0.04)	292 (21)	18.54 (0.05)	14.44	0.897
P50S50	50/50	w/o	Peat-Mineral-Mix	7.63 (0.03)	282 (32)	6.25 (0.11)	9.12	0.464
		w(+)		7.94 (0.02)	302 (33)	6.70 (0.06)	9.22	0.630
P30S70	33.3/66.7	w/o	Peat-Mineral-Mix	7.69 (0.05)	172 (13)	3.23 (0.06)	5.60	0.311
		w(+)		7.96 (0.03)	256 (29)	3.87 (0.05)	5.73	0.482
S100	0/100	w/o	Mineral Subsoil	6.89 (0.08)	87 (10)	0.70 (0.06)	0.31 (0.06)	0.006 (0.003)
		w(+)		8.03 (0.10)	371 (33)	0.99 (0.02)	0.50 (0.09)	0.186 (0.004)
FFM	NA	w/o	Forest-Floor-Mineral-Mix	6.77 (0.03)	161 (10)	4.09 (0.06)	2.09 (0.02)	0.074 (0.001)

Note: Treatment names are based on volumetric Peat to Subsoil ratios. Biochar was amended to Peat / Mineral Subsoil mix treatments at a rate of 10MT/ha (32.2g per pot) (w/o =no biochar added; w(+) = biochar added), pH (aq) and EC (µs/cm) reported as mean (standard error) n=9. %SOM based on loss on ignition n=3, expressed as percent based on weight loss. Total Carbon (TC) and Total Nitrogen (TN) n=3 based on combustion analysis for P100, S100, and FFM, weighted for admixing treatments based on S100 and P100 values. Treatment names indicate approximate admix ratios (e.g. 88.9 parts peat and 11.1 parts subsoil = P90S10).

Table 2: Comparison of mean value (±SE) by treatment effects on biometrics, selected foliar element conc., selected bioavailable nutrients (PRS™), and SOM Stability.

	Biometrics		**Foliar**									**PRS™**						**SOM**
Treatment	Biomass	Height	N	P	K	S	Mg	Ca	Na	Mn	Cu	N	P	K	S	Mg	Ca	Stability
	g	cm	mg g^{-1}							µg g^{-1}		µg $10cm^2$ 7 days						ratio
P100	0.94*c*	11.8*b*	25.15*a*	0.42*c*	13.44*ab*	2.85*ab*	3.47*b*	15.52*b*	0.45*ab*	44.5*c*	5.9*ab*	136.53*a*	0.36*b*	28.02*b*	312.91*a*	152.11*a*	2588.22*a*	0.40*ab*
	(0.16)	(1.6)	(0.89)	(0.02)	(0.71)	(0.22)	(0.22)	(1.35)	(0.01)	(2.0)	(1.4)	(32.32)	(0.15)	(8.05)	(32.32)	(18.35)	(262.83)	(0.01)
P90S10	0.77*c*	10.3*b*	27.51*a*	0.47*c*	16.25*a*	2.96*ab*	3.37*b*	15.25*b*	0.56*a*	55.2*c*	6.5*ab*	188.35*a*	0.06*b*	35.09*ab*	351.39*a*	133.53*a*	2337.17*a*	0.40*ab*
	(0.09)	(1.0)	(1.36)	(0.01)	(1.73)	(0.37)	(0.22)	(0.79)	(0.09)	(4.6)	(0.7)	(7.95)	(0.02)	(6.2)	(7.95)	(17.71)	(310.83)	(0.01)
P80S20	0.73*c*	7.8*b*	25.44*a*	0.55*c*	11.91*ab*	4.21*a*	4.38*b*	21.46*ab*	0.58*a*	85.2*c*	9.0*ab*	123.37*a*	0.21*b*	17.65*b*	324.57*a*	142.90*a*	2671.21*a*	0.43*a*
	(0.13)	(0.8)	(1.96)	(0.03)	(1.44)	(0.99)	(0.53)	(2.89)	(0.11)	(17.6)	(1.8)	(30.37)	(0.09)	(3.45)	(30.37)	(4.33)	(156.27)	(0.01)
P50S50	2.10*ab*	12.6*b*	16.46*b*	0.76*bc*	13.27*ab*	1.92*b*	3.77*b*	20.88*ab*	0.19*bc*	83.9*c*	4.8*b*	22.92*b*	0.15*b*	20.25*b*	137.84*b*	54.30*b*	899.38*b*	0.29*c*
	(0.52)	(2.4)	(1.76)	(0.05)	(1.16)	(0.16)	(0.29)	(2.81)	(0.04)	(8.1)	(1.4)	(6.06)	(0.05)	(0.73)	(6.06)	(3.00)	(41.63)	(0.04)
P30S70	0.97*bc*	7.7*b*	15.93*b*	1.03*b*	9.90*bc*	2.04*b*	5.01*b*	24.78*a*	0.27*bc*	140.4*b*	4.6*b*	13.17*b*	0.42*b*	25.60*b*	79.34*b*	68.71*b*	745.65*b*	0.27*c*
	(0.11)	(1.1)	(0.88)	(0.09)	(0.97)	(0.11)	(0.58)	(2.69)	(0.03)	(18.8)	(1.1)	(3.50)	(0.04)	(1.71)	(3.50)	(17.28)	(157.90)	(0.02)
S100	1.10*bc*	7.9*b*	14.41*b*	1.63*a*	6.21*c*	1.94*b*	7.27*a*	21.93*ab*	0.36*abc*	259.6*a*	7.7*ab*	13.05*b*	0.46*b*	15.87*b*	13.11*b*	23.98*b*	129.75*b*	0.15*d*
	(0.12)	(0.5)	(0.81)	(0.17)	(0.6)	(0.21)	(0.42)	(0.83)	(0.07)	(9.0)	(0.8)	(5.28)	(0.25)	(6.51)	(5.28)	(6.57)	(38.98)	(0.02)
FFM	2.65*a*	23.6*a*	18.88*b*	1.72*a*	12.27*ab*	1.84*b*	4.40*b*	18.80*ab*	0.10*c*	175.6*b*	11.3*a*	35.31*b*	2.50*a*	59.92*a*	64.95*b*	69.71*b*	607.58*b*	0.33*bc*
	(0.35)	(3.0)	(1.38)	(0.10)	(0.93)	(0.10)	(0.39)	(1.50)	(0.02)	(11.4)	(1.6)	(4.48)	(0.22)	(8.68)	(4.48)	(9.23)	(52.87)	(0.02)

Note: Foliar element conc. (n=6), selected bioavailable nutrients (PRS™) (n=3), and SOM Stability (n=9). tested with permutational ANOVA and TUKEY HSD as a posthoc test with alpha=0.05. Different letters indicate significant differences within columns only.

Table 3: Comparison of mean value (±SE) by treatment effects on biometrics, selected foliar element conc., selected bioavailable nutrients (PRS™), and SOM Stability.

	Biometrics		Foliar									PRS™						SOM
Treatment	Biomass	Height	N	P	K	S	Mg	Ca	Na	Mn	Cu	N	P	K	S	Mg	Ca	Stability
	g	cm	mg g^{-1}							µg g^{-1}		µg 10cm^2 7 days						ratio
P100+	1.19*b*	11.4*b*	22.65*a*	0.53*c*	11.77*ab*	2.33*a*	3.32*a*	16.87*b*	0.34*ab*	55.9*b*	5.7*ab*	193.48*a*	0.36*b*	28.96*b*	383.71*a*	128.25*ab*	2110.43*ab*	0.41*a*
	(0.29)	(2.1)	(1.19)	(0.05)	(1.41)	(0.18)	(0.44)	(1.91)	(0.05)	(9.37)	(0.81)	(78.76)	(0.16)	(3.69)	(199.18)	(7.95)	(87.97)	(0.01)
P90S10+	1.39*b*	13.6*b*	20.99*ab*	0.47*c*	12.62*a*	2.54*a*	3.51*a*	18.38*b*	0.41*ab*	55.1*b*	5.6*ab*	98.05*ab*	0.26*b*	29.11*b*	378.63*a*	135.79*ab*	2328.16*a*	0.41*a*
	(0.31)	(2.8)	(1.86)	(0.04)	(0.51)	(0.49)	(0.60)	(2.29)	(0.10)	(11.32)	(2.09)	(14.94)	(0.09)	(2.38)	(113.47)	(12.35)	(209.33)	(0.01)
P80S20+	1.34*b*	13.7*ab*	20.05*ab*	0.64*c*	13.36*a*	2.22*a*	3.41*a*	17.03*b*	0.28*ab*	50.9*b*	4.9*b*	64.17*ab*	0.17*b*	28.16*b*	351.36*a*	144.58*a*	2448.18*a*	0.43*a*
	(0.35)	(2.6)	(1.65)	(0.16)	(1.01)	(0.06)	(0.56)	(2.47)	(0.09)	(13.42)	(0.65)	(7.47)	(0.15)	(2.07)	(70.90)	(22.99)	(439.05)	(0.01)
P50S50+	1.52*ab*	13.2*b*	16.01*bcd*	1.31*ab*	11.38*ab*	2.12*a*	4.62*a*	29.70*a*	0.20*b*	74.5*b*	3.9*b*	43.13*b*	0.47*b*	32.46*b*	385.34*a*	111.58*ab*	1892.16*ab*	0.33*b*
	(0.2)	(0.8)	(0.64)	(0.18)	(0.96)	(0.10)	(0.42)	(2.05)	(0.02)	(7.13)	(0.60)	(7.14)	(0.21)	(2.47)	(66.82)	(11.52)	(61.06)	(0.02)
P30S70+	1.37*b*	12.3*b*	13.47*cd*	0.81*bc*	10.61*ab*	2.13*a*	3.39*a*	16.93*b*	0.27*ab*	73.5*b*	4.5*b*	12.93*b*	0.73*b*	33.06*b*	182.71*a*	86.29*abc*	1203.73*bc*	0.22*c*
	(0.21)	(2.3)	(1.41)	(0.16)	(0.47)	(0.21)	(0.31)	(1.94)	(0.05)	(14.90)	(1.75)	(2.43)	(0.40)	(1.48)	(35.48)	(19.74)	(201.99)	(0.01)
S100+	0.79*b*	8.0*b*	12.55*d*	1.02*bc*	8.37*b*	2.96*a*	4.30*a*	18.68*b*	0.62*a*	86.5*b*	6.1*ab*	7.51*b*	0.89*b*	29.31*b*	86.26*a*	34.65*c*	448.39*c*	0.17*c*
	(0.2)	(1.3)	(0.94)	(0.13)	(1.03)	(1.21)	(0.35)	(2.48)	(0.19)	(15.05)	(1.18)	(1.39)	(0.14)	(2.36)	(18.02)	(6.29)	(67.84)	(0.02)
FFM	2.65*a*	23.6*a*	18.88*abc*	1.72*a*	12.27*ab*	1.84*a*	4.40*a*	18.80*b*	0.10*b*	175.6*a*	11.3*a*	35.31*b*	2.5*a*	59.92*a*	64.95*a*	69.71*bc*	607.58*c*	0.33*b*
	(0.35)	(3.0)	(1.38)	(0.10)	(0.93)	(0.10)	(0.39)	(1.50)	(0.02)	(11.43)	(1.64)	(4.48)	(0.22)	(8.68)	(4.48)	(9.23)	(52.87)	(0.02)

Note: Foliar element conc. (n=6), selected bioavailable nutrients (PRS™) (n=3), and SOM Stability (n=9). tested with permutational ANOVA and TUKEY HSD as a posthoc test with alpha=0.05. Different letters indicate significant differences within columns only. Treatments with a plus symbol (+) indicate biochar amendment with a rate of 10 MT/ha.

List of Figures

Figure 1: Principal Component Analysis (PCA) of standardized foliar nutrient concentration from aspen seedlings (n=6) with no biochar amendment. For treatment identification, see Table 1. Colored ellipses represent 68 percent confidence interval for each treatment.

Figure 2: Principal Component Analysis (PCA) of standardized bioavailable nutrients (PRS™) (n=3) and soil organic matter stability (Q2/Q1 ratio) (n=3) in pots with no tree and no biochar amendment. For treatment, identification see Table 1. Colored ellipses represent the 68 percent confidence interval for each treatment.

Figure 3: Principal Component Analysis (PCA) of standardized bioavailable nutrients (PRS™) (n=3), soil organic matter stability (Q2/Q1 ratio) (n=3) in pots with no tree and with and without biochar amendment, and equivalent averaged foliar element concentrations and biometric characteristics (biomass, height). Treatments marked with (+) indicate biochar amendment at a rate of 10MT/ha. Colored ellipses represent 68 percent confidence interval for each treatment. Arrows indicate a trend for a shift with biochar amendment for treatments P 100, P90S10, P80S20 (left arrow) and P30S70, and S100 (right arrow). Arrows are added for graphical purpose only.

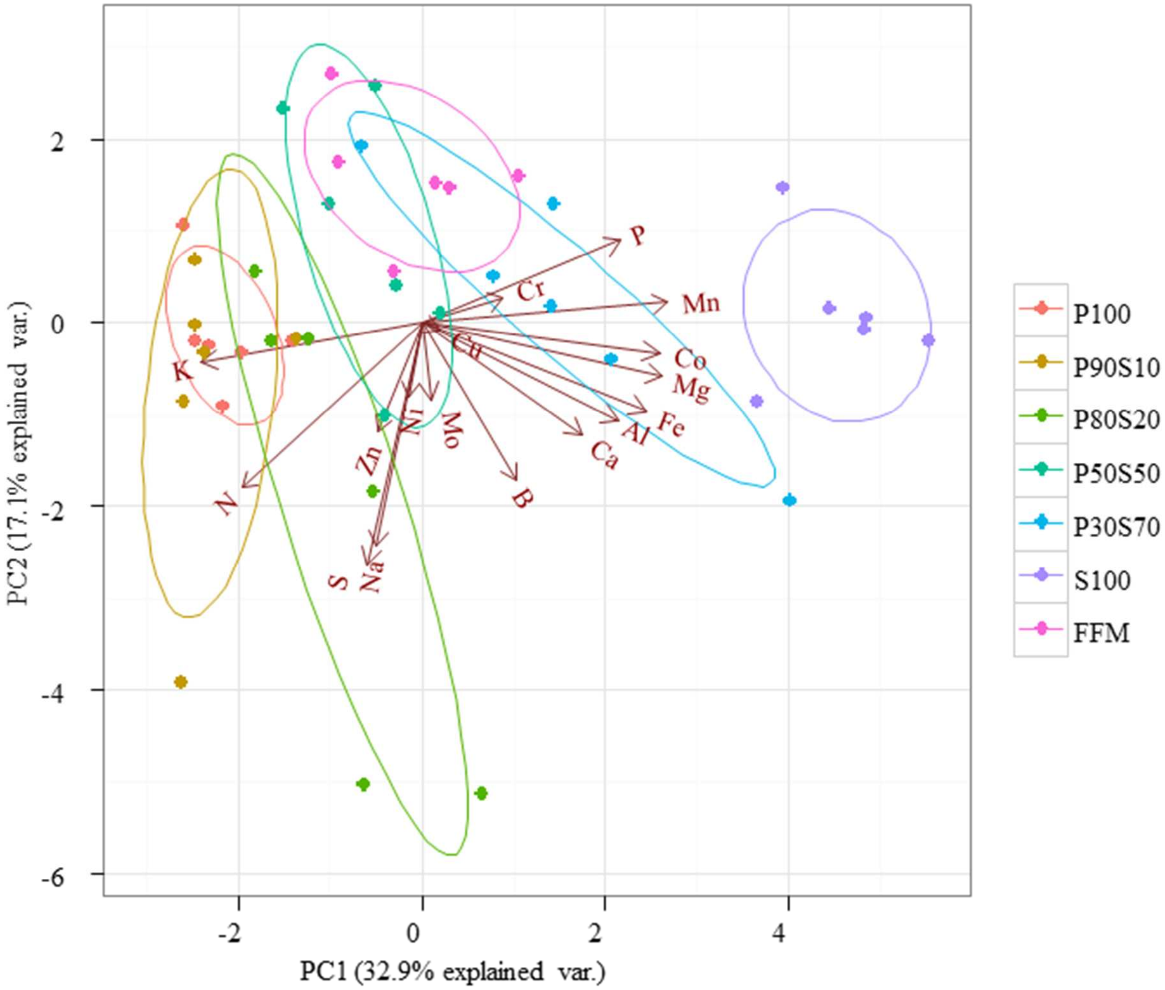
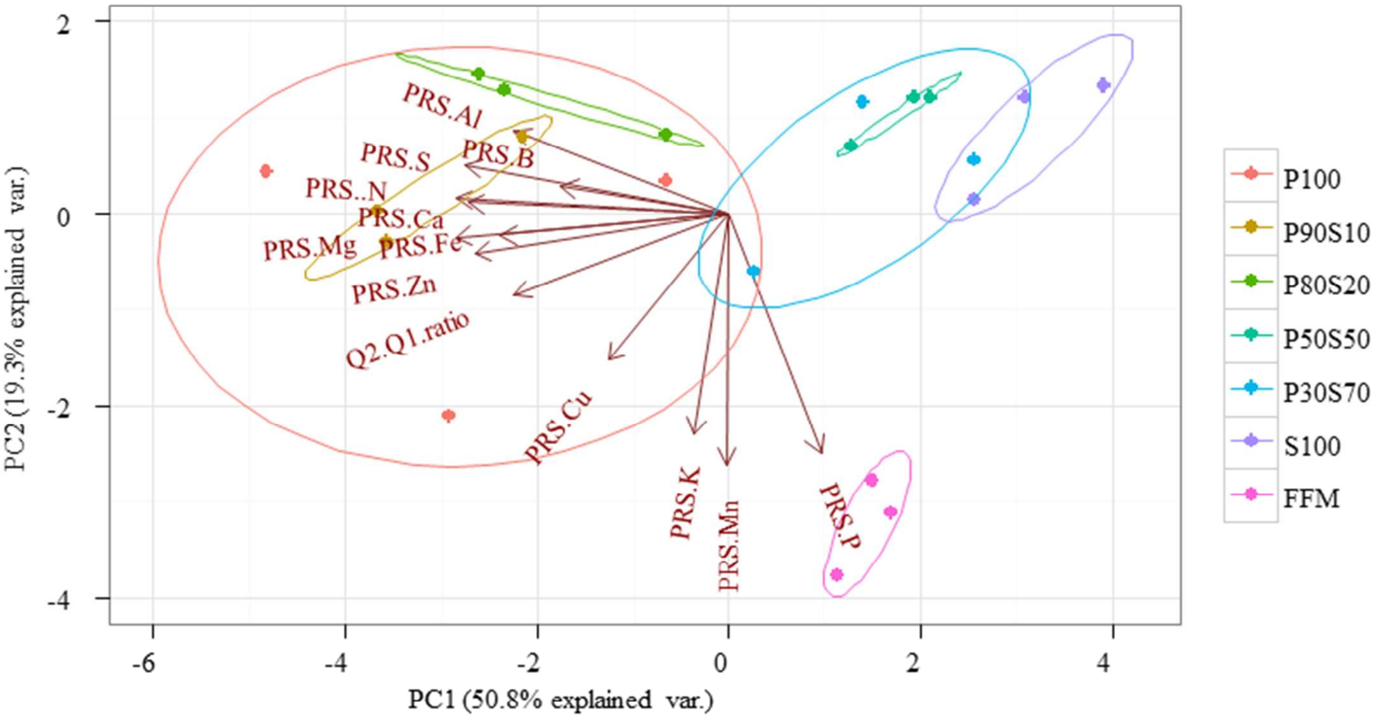

PC2 (17.1% explained var.)
PC1 (32.9% explained var.)
2
0
-2
-4
-6
-2
0
2
4
P
Cr
Mn
Cu
Co
Mg
Fe
Al
Ca
B
Mo
Ni
Zn
S
Na
N
K
P100
P90S10
P80S20
P50S50
P30S70
S100
FFM

Figure 1



2
0
-2
-4
-6
-4
-2
0
2
4
PC2 (19.3% explained var.)
PC1 (50.8% explained var.)
PRS.Al
PRS.S
PRS.B
PRS..N
PRS.Ca
PRS.Mg
PRS.Fe
PRS.Zn
Q2.Q1.ratio
PRS.Cu
PRS.K
PRS.Mn
PRS.P
P100
P90S10
P80S20
P50S50
P30S70
S100
FFM

Figure 1

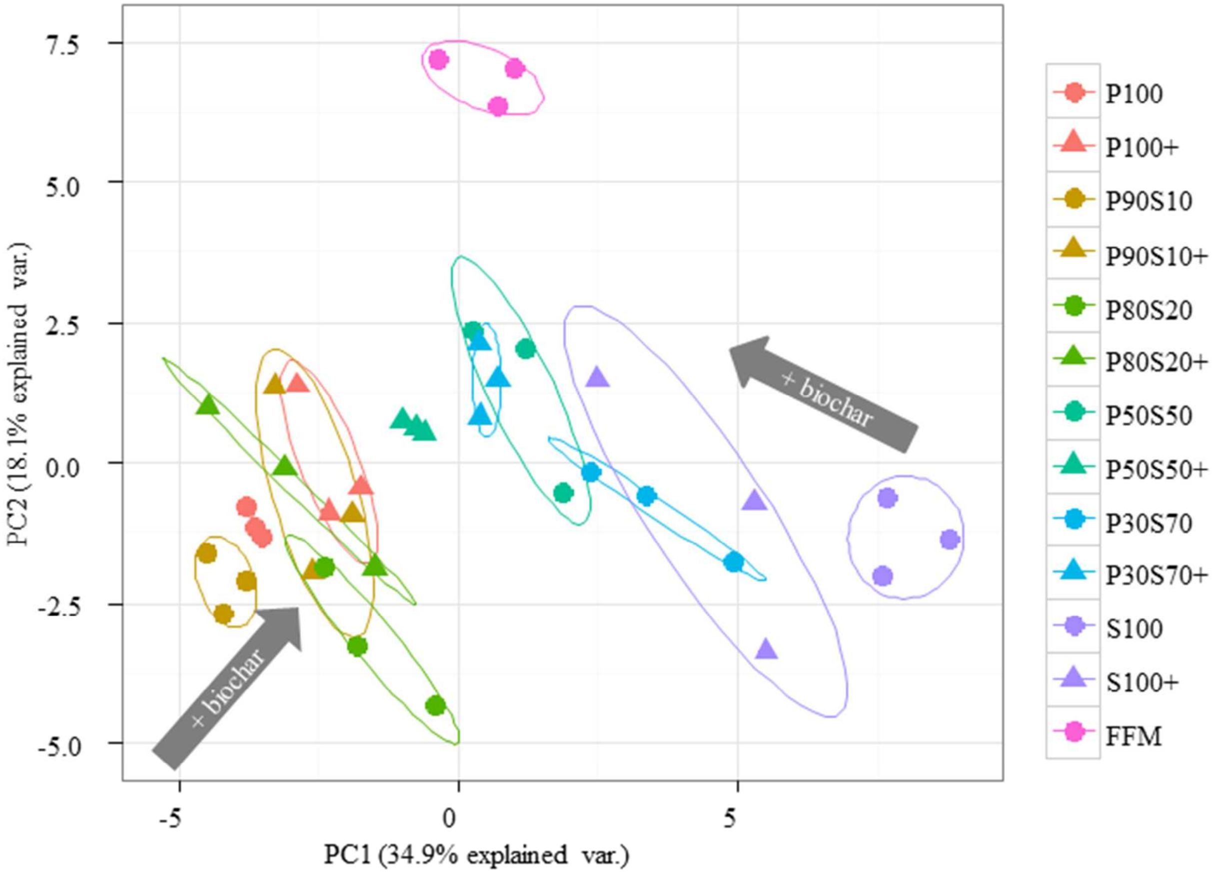

7.5
5.0
2.5
0.0
-2.5
-5.0
PC2 (18.1% explained var.)
-5
0
5
PC1 (34.9% explained var.)
+ biochar
+ biochar
P100
P100+
P90S10
P90S10+
P80S20
P80S20+
P50S50
P50S50+
P30S70
P30S70+
S100
S100+
FFM

Figure 2